

Docket No.: 20347 US1 (C38435/128985)

September 30, 2002

## IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

For: β,β-CAROTENE 15,15'- MONOOXYGENASES, NUCLEIC ACID SEQUENCES CODING THEREFOR AND THEIR USE	) New York, NY			
Filed: January 15, 2002	)			
Serial No.: 10/053,192	, ,			
Heinrich BACHMANN, et al.	BACHMANN, et al.  ) Examiner: not yet assigned			
n re Application of :	ation of :			

## TRANSMITTAL OF PROPOSED DRAWING CORRECTION

Box Missing Parts Commissioner For Patents Washington, D.C. 20231 Sir:

In accordance with 37 CFR § 121(d) and MPEP § 608.02(q) and at the request of the Examiner, (see Notice to Filing Missing Parts mailed April 29, 2002), the attached corrected drawing figures (see Exhibit 1) are submitted for consideration. It is respectfully requested that the Examiner enter corrected figures 2-8 in this application.

The correction to figures 2-5 is as follows:

a. figures 2-5 have been reformatted to correct the sizes of the margins in order to conform to the requirements of 37 CFR § 1.84(g).

The corrections to figures 6-8 are as follows:

- a. figures 7 and 8 have been reformatted to correct the sizes of the margins in order to conform to the requirements of 37 CFR § 1.84(g).
- b. all descriptive text, other than text indicating the figure number, has been cancelled from figures 6-8 in order to conform to the requirements of 37 CFR § 1.84(o).

For the convenience of the Examiner, we attach as Exhibit 2 a copy of figures 6-8 as amended (without the redline).

It is submitted that the entry of the corrections to Figures 2-8 does not introduce new matter, and is proper. See 37 CFR § 121(a)(6). Accordingly, entry of the attached corrected drawing figures in the above referenced application, respectfully is requested. If the Examiner has any questions regarding this paper, please contact the undersigned.

I hereby certify that this correspondence is being deposited with the United States Postal Service with sufficient postage as first class mail in an envelope addressed to Box Missing Parts, Commissioner For Patents, Washington, D.C. 20231, on September 30, 2002.

Gonzalo Merino

Respectfully submitted,

Gonzalo Merino

Registration No. 51,192

BRYAN CAVE LLP

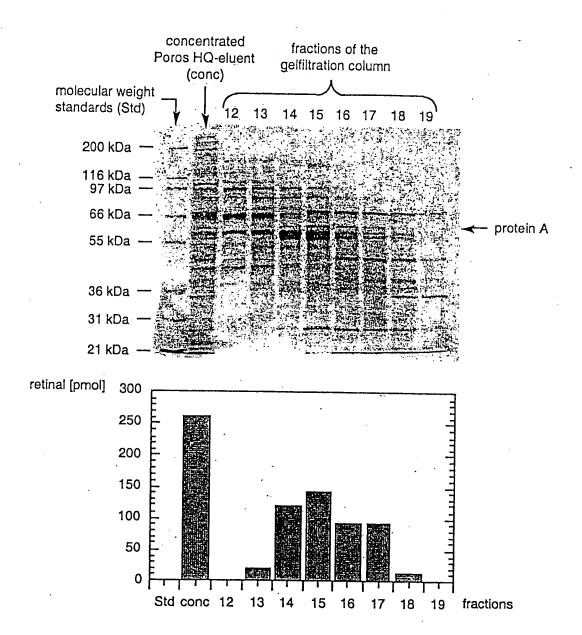
245 Park Avenue

New York, NY 10167-0034

(212) 692-1800



Figure 1





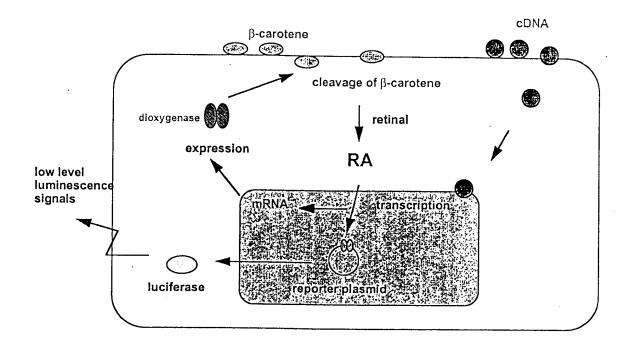


Figure 2



Figure 3 No. 2

- 1 CGGATCCACT AGTAACGGCC GCCAGTGTGG TGGAATCCAT
- 51 AACAGGAAAG AGCTGTTCTT AGCCCAGAGA GGAGGGCACC GTACGCCTGC
- 101 AGGAGCAGCT GGGTAGAGGA CACAGGAGAG CGATGGAGAC AATATTTAAC
- 151 AGAAACAAAG AAGAGCATCC AGAGCCCATA AAAGCTGAGG
- 201 GTTGCCCACT TGGTTGCAAG GGGTACTTCT CCGAAATGGC CCAGGGATGC
- 251 ACACAATAGG GGACACTAAA TACAACCACT GGTTTGATGG
- 301 CTGCACAGCT TCACGTTTAA AAATGGTGAA GTTTACTACA GAAGTAAGTA
- 351 CCTCCGAAGT GACACATACA ACTGCAATAT AGAAGCAAAC CGAATCGTGG
- 401 TGTCTGAGTT TGGAACCATG GCTTATCCGG ATCCATGCAA AAACATATTT ...
- 451 GCCAAGGCAT TCTCATACTT ATCTCACACC ATTCCTGAGT TCACGGACAA
- 501 CTGCCTGATC AACATTATGA AAACTGGGGA TGATTATTAT GCTACCAGTG
- 551 AGACTAACTT CATCAGAAAA ATTGATCCAC AGACTCTGGA GACACTAGAT
- 601 AAGGTAGACT ACAGCAAATA TGTAGCTGTA AACTTGGCAA
- 651 ACACTATGAC AGTGCTGGAA ATATTCTCAA CATGGGTACT TCAATTGTTG
- 701 ATAAAGGGAG AACAAAATAT GTTCTCTTTA AGATCCCTTCCTCTGTACCA
- 751 GAAAAAGAAA AGAAGAAATC TTGTTTTAAA CACCTGGAAG TAGTATGCTC
- 801 CATCCCTTCT CGCTCCCTGC TCCAACCAAG CTACTACCAC AGCTTTGGAA
- 851 TCACAGAAAA TTATATTGTG TTCATAGAGC AGCCATTTAA ACTGGATATT

- 901 GTCAAACTGG CAACTGCCTA CATCCGAGGT GTGAACTGGG
- 951 TTCCTTTCAT AAGGAGGATA AGACGTGGTT TCACTTTGTA GACAGAAAGA
- 1001 CGAAAAAAGA AGTATCCACC AAGTTTTACA CTGATGCTTT GGTGCTTTAT
- 1051 CACCACATAA ATGCTTACGA AGAAGATGGC CACGTTGTTT TTGATATCGT
- 1101 TGCCTACAGA GACAATAGCT TGTACGATAT GTTTTACTTA AAAAAACTGG
- 1151 ACAAAGACTT TGAAGTGAAC AACAAGCTTA CCTCCATCCC AACCTGCAAG
- 1201 CGCTTTGTTG TGCCTCTGCA GTATGACAAG GATGCAGAAG TAGGTTCTAA
- 1251 TTTAGTCAAA CTTCCAACTT CCGCAACTGC TGTAAAAGAA AAAGATGGCA
- 1301 GCATCTATTG TCAACCTGAA ATATTATGTG AAGGGATAGA ACTGCCTCGT
- 1351 GTCAACTATG ACTACAATGG CAAAAAATAC AAGTATGTCT ATGCAACAGA
- 1401 AGTCCAGTGG AGCCCAGTTC CTACAAAGAT TGCAAAACTG AATGTCCAAA
- 1451 CAAAGGAAGT ACTGCACTGG GGAGAAGACC ACTGCTGGCC CTCAGAGCCC
- 1501 ATCTTTGTTC CCAGCCCCGA TGCAAGAGAA GAGGATGAAG GTGTTGTTTT
- 1551 GACCTGTGTT GTGGTGTCTG AGCCAAATAA AGCACCCTTC CTACTCATCT
- 1601 TGGATGCTAA AACATTCAAA GAATTGGGCC GAGCCACAGT TAACGTAGAA
- 1651 ATGCATCTGG ACCTGCATGG GATGTTTATA CCACAGAATG ATTTGGGGGC
- 1701 TGAGACGGAA TAAAACGCTA TTGATCCGAC TACACAAACT GAGACAACTT
- 1751 TCTACTGAAC ATGAGTTAAT ATCCCTTTTA CCATTCAAGA ACAACCATAT
- 1801 AACGACACAA AATGACTATG TATAATCTCT TAAATAATAG ATATAATCCT
- 1851 TTTAAGGCAC AGCGATGAGT TTTACTACAG GTAACGATAT GCACAACTGG



- 1901 CATATAACTA TTCCAAAAGA AGAAGAACGA TCAGTGTTTT AGAAGTGCTA
- 1951 ATGTTGTACA TAACGGCGGC AGAGGGAACA GGAGAGAAAG GTAACGGAA
- 2001 TATTTAATAG AATATAGATT TCTGAGCAAA TGAAGTGCAG TATTTATGGT
- 2051 GTGATGCATG GCATGAGTCA CATAGGTCTG CAGCTCATGT ATCTTTTAGA
- 2101 GATCGTTTCA AGATTGCAGC TTGTGATGCA AGTTTTCTCC AGCCAGAAAA
- 2151 CCTCATTTTA AACCATCTGC TACTGGTAAT TCATACCAAT GCATTTTCTT
- 2201 GGTGCTCGAT TTACACTATA ACCAAAGTTA AGTATTACAT TCAGGTGCTA
- 2251 CAACTTTCTA ATTTACAACC GAAACAACA AGCAAACAGC ACTTGCTTTG
- 2301 CTAATAACCC CATGGTGTAT TTTTCCTTTT TATGATGACA AAACCAAGTA
- 2351 CATATGGTTT TATGTAGCAT TCAATTATAC TTCAGTGCTA
- 2401 ATGTTATAAG CAATTTGTAT TTAAATCAGT TTTCCTTGAG AATATCTGAC
- 2451 ATAACATTTT GTGTAATGAG ATGACTATGT TGTCTAAAGA TGAACAGGAA
- 2501 TGTATCTTTT ATTAGTATTG TTAATTGTGT TACTAATACT ATGCATATGA
- 2551 ATGAGAGCAA TGTATTTCTA GGAGAACTCA GATATACATT CAACAATTTC
- 2601 TGTAGGTGAA AATGCATTTA CTGATGAAAG TTGAATCGTT AATGAGGGAG
- 2651 AAAACTGGGT ATCCATCCAT CCAACTATGT TAGGTGTTCA
- 2701 ATGTGACACC ACGCTGTTTG GGTATCTCTC ACTTTCACAT ACCTGTTCTC
- 2751 ATGGTTTCTG CTACTCACTG TATTTTGCAG GAGAGAAACA AAATGAAATC
- 2801 ACTGTCACTT ACTATCGCCC CATCACATAA GAACAATGGG GCTTTGGTGA

- 2851 CTTGTTCATG ATTACATAAG ATGTTTGCAG CAGAGCAGCA ATAGAACCAA
- 2901 CACCATCCAC AGTTCTTGCT TGCTCTGTTA TGACTCCCTT TGCTGTCTTT
- 2951 ATGGTTTGCA TGTATGAAGA ATACACTGCC TAATTCTAAT GTTAAAAAGT
- 3001 CACTGGGGTC AGATCTAGAG CTTAAGTAAG CAGTCTGGGG
- - 3101 AAAAAAAAA A

Figure 4 No. 1

Seq. ID

- 1 METIFNRNKE EHPEPIKAEV QGQLPTWLQG VLLRNGPGMH TIGDTKYNHW
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- 101 PCKNIFAKAF SYLSHTIPEF TDNCLINIMK TGDDYYATSE TNFIRKIDPQ
- 151 TLETLDKVDY SKYVAVNLAT SHPHYDSAGN ILNMGTSIVD KGRTKYVLFK
- 201 IPSSVPEKEK KKSCFKHLEV VCSIPSRSLL QPSYYHSFGI TENYIVFIEQ
- 251 PFKLDIVKLA TAYIRGVNWA SCLSFHKEDK TWFHFVDRKT KKEVSTKFYT
- 301 DALVLYHHIN AYEEDGHVVF DIVAYRDNSL YDMFYLKKLD KDFEVNNKLT
- 351 SIPTCKRFVV PLQYDKDAEV GSNLVKLPTS ATAVKEKDGS IYCOPEILCE
- 401 GIELPRVNYD YNGKKYKYVY ATEVQWSPVP TKIAKLNVQT KEVLHWGEDH ...
- 451 CWPSEPIFVP SPDAREEDEG VVLTCVVVSE PNKAPFLLIL DAKTFKELGR
  - 501 ATVNVEMHLD LHGMFIPQND LGAETE

Figure 5

Seq ID No. 4 and Seq ID No. 5

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105	58	HSFTFKNGEVYYRSKYLRSDTYNCNIEANRIVVSEFGTMAYPDPCKNI
119	70	::: .  .    :.      :       HKFDFKEGHVTYHRRFIRTDAYVRAMTEKRIVITEFGFTTCAFPDPCKNI
155		FAKAFSYLSHTIPEFTDNCLINIMKTGDDYYATSETNFIRKIDPQTLETI
167	120	.:            : :  :     .        : :    :   :
203		. DKVDYSKYVAVNLATSHPHYDSAGNILNMGTSIVDKGRTKYVLFKIPS
217	168	.     .     .   :.   :  .    :     FTKQVDLCNYVSVNGATAHPHIENDGTVYNIGNCFGKNFSIAYNIVKIPF
253		SVPEKEKKKSCFKHLEVVCSIPSRSLLQPSYYHSFGITENYIVFIEQPFK
266	218	:
300	254	LDIVKLATAY.IRGVNWASCL.SFHKEDK.TWFHFVDRKTKKEVSTKFYT
316	267	:.:  : :    :
344	301	DALVLYHHINAYEEDGHVVFDIVAYRDNSLYDMFYLKKLDKDFE
366	317	:      :.  .:  : ::            SPFNLFHHINTYEDNGFLIVDLCCWKGFEFVYNYFTLYLANLRENWEEVK
391		VNNKLTSIPTCKRFVVPLQYDKDAEVGSNLVKLP.TSATAVKEKDGSI
115	367	:  : .       :                     KNARKAPQPEVRRYVLPLNIDK.ADTGKNLVTLPNTTATAILCSDEFTTI
136		YCQPEILCEGIELPRVNYD.YNGKKYKYVYATEVQWSPVPTKIAKL
164	416	::  :



484	437	NVQTKEVLHWGEDHCWPSEPIFVPSPDAREEDEGVVLTCVVVSEPNKA
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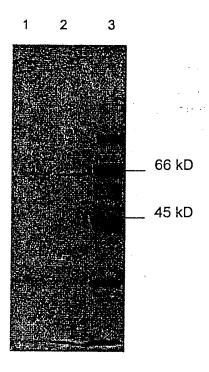


Fig. 6



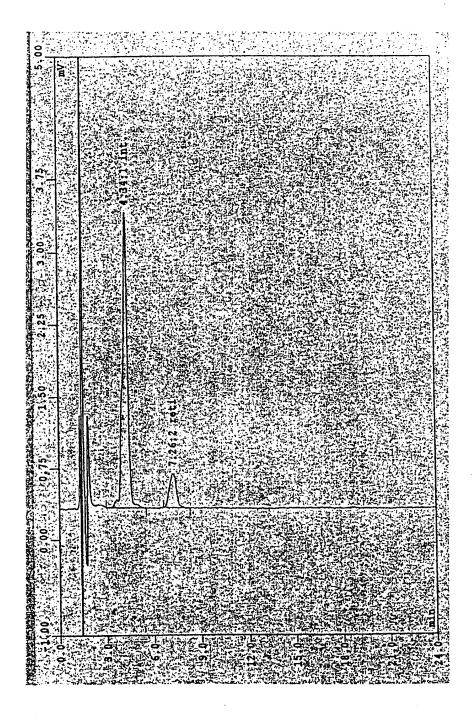


Fig. 7



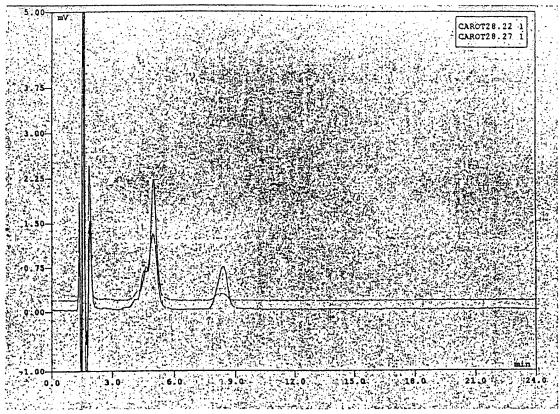
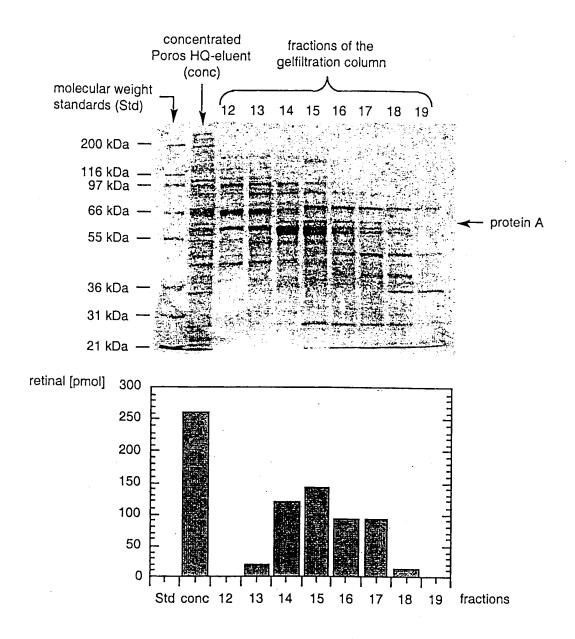


Fig. 8



Figure 1





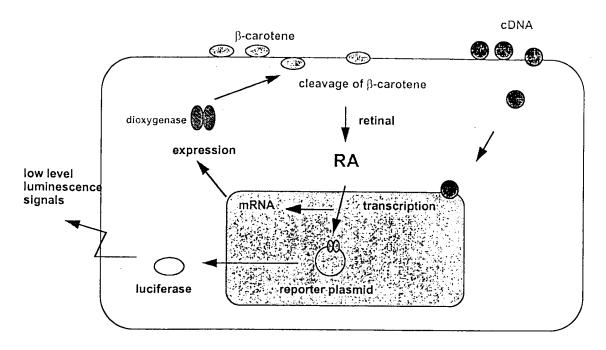


Figure 2



Figure 3 No. 2

- 1 CGGATCCACT AGTAACGGCC GCCAGTGTGG TGGAATCCAT
- 51 AACAGGAAAG AGCTGTTCTT AGCCCAGAGA GGAGGGCACC GTACGCCTGC
- 101 AGGAGCAGCT GGGTAGAGGA CACAGGAGAG CGATGGAGAC AATATTTAAC
- 151 AGAAACAAAG AAGAGCATCC AGAGCCCATA AAAGCTGAGG TGCAAGGTCA
- 201 GTTGCCCACT TGGTTGCAAG GGGTACTTCT CCGAAATGGC CCAGGGATGC
- 251 ACACAATAGG GGACACTAAA TACAACCACT GGTTTGATGG
- 301 CTGCACAGCT TCACGTTTAA AAATGGTGAA GTTTACTACA GAAGTAAGTA
- 351 CCTCCGAAGT GACACATACA ACTGCAATAT AGAAGCAAAC CGAATCGTGG
- 401 TGTCTGAGTT TGGAACCATG GCTTATCCGG ATCCATGCAA AAACATATTT
- 451 GCCAAGGCAT TCTCATACTT ATCTCACACC ATTCCTGAGT TCACGGACAA
- 501 CTGCCTGATC AACATTATGA AAACTGGGGA TGATTATTAT GCTACCAGTG
- 551 AGACTAACTT CATCAGAAAA ATTGATCCAC AGACTCTGGA GACACTAGAT
- 601 AAGGTAGACT ACAGCAAATA TGTAGCTGTA AACTTGGCAA
- 651 ACACTATGAC AGTGCTGGAA ATATTCTCAA CATGGGTACT TCAATTGTTG
- 701 ATAAAGGGAG AACAAAATAT GTTCTCTTTA AGATCCCTTC
- 751 GAAAAAGAAA AGAAGAAATC TTGTTTTAAA CACCTGGAAG TAGTATGCTC
- 801 CATCCCTTCT CGCTCCCTGC TCCAACCAAG CTACTACCAC AGCTTTGGAA
- 851 TCACAGAAAA TTATATTGTG TTCATAGAGC AGCCATTTAA ACTGGATATT



- 901 GTCAAACTGG CAACTGCCTA CATCCGAGGT GTGAACTGGG
- 951 TTCCTTTCAT AAGGAGGATA AGACGTGGTT TCACTTTGTA GACAGAAAGA
- 1001 CGAAAAAGA AGTATCCACC AAGTTTTACA CTGATGCTTT GGTGCTTTAT
- 1051 CACCACATAA ATGCTTACGA AGAAGATGGC CACGTTGTTT TTGATATCGT
- 1101 TGCCTACAGA GACAATAGCT TGTACGATAT GTTTTACTTA
- 1151 ACAAAGACTT TGAAGTGAAC AACAAGCTTA CCTCCATCCC AACCTGCAAG
- 1201 CGCTTTGTTG TGCCTCTGCA GTATGACAAG GATGCAGAAG TAGGTTCTAA
- 1251 TTTAGTCAAA CTTCCAACTT CCGCAACTGC TGTAAAAGAA AAAGATGGCA
- 1301 GCATCTATTG TCAACCTGAA ATATTATGTG AAGGGATAGA ACTGCCTCGT
- 1351 GTCAACTATG ACTACAATGG CAAAAAATAC AAGTATGTCT ATGCAACAGA
- 1401 AGTCCAGTGG AGCCCAGTTC CTACAAAGAT TGCAAAACTG
- 1451 CAAAGGAAGT ACTGCACTGG GGAGAAGACC ACTGCTGGCC CTCAGAGCCC
- 1501 ATCTTTGTTC CCAGCCCCGA TGCAAGAGAA GAGGATGAAG GTGTTGTTTT
- 1551 GACCTGTGTT GTGGTGTCTG AGCCAAATAA AGCACCCTTC CTACTCATCT
- 1601 TGGATGCTAA AACATTCAAA GAATTGGGCC GAGCCACAGT TAACGTAGAA
- 1651 ATGCATCTGG ACCTGCATGG GATGTTTATA CCACAGAATG ATTTGGGGGC
- 1701 TGAGACGGAA TAAAACGCTA TTGATCCGAC TACACAAACT GAGACAACTT
- 1751 TCTACTGAAC ATGAGTTAAT ATCCCTTTTA CCATTCAAGA ACAACCATAT
- 1801 AACGACACAA AATGACTATG TATAATCTCT TAAATAATAG ATATAATCCT
- 1851 TTTAAGGCAC AGCGATGAGT TTTACTACAG GTAACGATAT GCACAACTGG



- 1901 CATATAACTA TTCCAAAAGA AGAAGAACGA TCAGTGTTTT AGAAGTGCTA
- 1951 ATGTTGTACA TAACGGCGGC AGAGGGAACA GGAGAGAAAG GTAACGGGAA
- 2001 TATTTAATAG AATATAGATT TCTGAGCAAA TGAAGTGCAG TATTTATGGT
- $2051\,$  GTGATGCATG GCATGAGTCA CATAGGTCTG CAGCTCATGT ATCTTTTAGA
- 2101 GATCGTTTCA AGATTGCAGC TTGTGATGCA AGTTTTCTCC AGCCAGAAAA
- 2151 CCTCATTTA AACCATCTGC TACTGGTAAT TCATACCAAT GCATTTTCTT
- 2201 GGTGCTCGAT TTACACTATA ACCAAAGTTA AGTATTACAT TCAGGTGCTA
- 2251 CAACTTTCTA ATTTACAACC GAAACAAACA AGCAAACAGC ACTTGCTTTG
- 2301 CTAATAACCC CATGGTGTAT TTTTCCTTTT TATGATGACA AAACCAAGTA
- 2351 CATATGGTTT TATGTAGCAT TCAATTATAC TTCAGTGCTA TTCCATCCTA ...
- 2401 ATGTTATAAG CAATTTGTAT TTAAATCAGT TTTCCTTGAG AATATCTGAC
- 2451 ATAACATTTT GTGTAATGAG ATGACTATGT TGTCTAAAGA TGAACAGGAA
- 2501 TGTATCTTTT ATTAGTATTG TTAATTGTGT TACTAATACT ATGCATATGA
- 2551 ATGAGAGCAA TGTATTTCTA GGAGAACTCA GATATACATT CAACAATTTC
- 2601 TGTAGGTGAA AATGCATTTA CTGATGAAAG TTGAATCGTT AATGAGGGAG
- 2651 AAAACTGGGT ATCCATCCAT CCAACTATGT TAGGTGTTCA
- 2701 ATGTGACACC ACGCTGTTTG GGTATCTCTC ACTTTCACAT ACCTGTTCTC
- 2751 ATGGTTTCTG CTACTCACTG TATTTTGCAG GAGAGAAACA
- 2801 ACTGTCACTT ACTATCGCCC CATCACATAA GAACAATGGG GCTTTGGTGA

- 2851 CTTGTTCATG ATTACATAAG ATGTTTGCAG CAGAGCAGCA ATAGAACCAA
- 2901 CACCATCCAC AGTTCTTGCT TGCTCTGTTA TGACTCCCTT TGCTGTCTTT
- 2951 ATGGTTTGCA TGTATGAAGA ATACACTGCC TAATTCTAAT GTTAAAAAGT
- 3001 CACTGGGGTC AGATCTAGAG CTTAAGTAAG CAGTCTGGGG
- 3051 TTTATATGTT CCATAAAATG GAAATAAACA CCTCCATAAT AAAAAAAAA
  - 3101 AAAAAAAAA A



Figure 4 No. 1

Seq. ID

- 1 METIFNRNKE EHPEPIKAEV QGQLPTWLQG VLLRNGPGMH TIGDTKYNHW
- 51 FDGLALLHSF TFKNGEVYYR SKYLRSDTYN CNIEANRIVV SEFGTMAYPD
- 101 PCKNIFAKAF SYLSHTIPEF TDNCLINIMK TGDDYYATSE TNFIRKIDPQ
- 151 TLETLDKVDY SKYVAVNLAT SHPHYDSAGN ILNMGTSIVD KGRTKYVLFK
- 201 IPSSVPEKEK KKSCFKHLEV VCSIPSRSLL QPSYYHSFGI TENYIVFIEO
- 251 PFKLDIVKLA TAYIRGVNWA SCLSFHKEDK TWFHFVDRKT KKEVSTKFYT
- 301 DALVLYHHIN AYEEDGHVVF DIVAYRDNSL YDMFYLKKLD KDFEVNNKLT
- 351 SIPTCKRFVV PLQYDKDAEV GSNLVKLPTS ATAVKEKDGS IYCOPEILCE
- 401 GIELPRVNYD YNGKKYKYVY ATEVQWSPVP TKIAKLNVQT KEVLHWGEDH
- 451 CWPSEPIFVP SPDAREEDEG VVLTCVVVSE PNKAPFLLIL DAKTFKELGR
  - 501 ATVNVEMHLD LHGMFIPQND LGAETE

Figure 5

Seq ID No. 4 and Seq ID No. 5

57	10	EEHPEPIKAEVQGQLPTWLQGVLLRNGPGMHTIGDTKYNHWFDGLALI
69	20	:      .:              : :  :            EELSSPLTAHVTGRIPLWLTGSLLRCFTGPGLFEVGSEPFYHLFDGQALI
105	58	
119	70	::: .  .    :.       :       HKFDFKEGHVTYHRRFIRTDAYVRAMTEKRIVITEFGFTTCAFPDPCKN
155	106	FAKAFSYLSHTIPEFTDNCLINIMKTGDDYYATSETNFIRKIDPQTLETI
167	120	.:             : :  :    .
203	156	DKVDYSKYVAVNLATSHPHYDSAGNILNMGTSIVDKGRTKYVLFKIPS
217	168	.     .     .   :.   :  .  FTKQVDLCNYVSVNGATAHPHIENDGTVYNIGNCFGKNFSIAYNIVKIP
253	204	SVPEKEKKKSCFKHLEVVCSIPSRSLLQPSYYHSFGITENYIVFIEQPF
266	218	:
300	254	LDIVKLATAY.IRGVNWASCL.SFHKEDK.TWFHFVDRKTKKEVSTKFYT
316	267	:.:  : :    :               :      :   INLFKFLSSWSLWGANYMDCFESFTNETMGVWLHIADKKRKKYLNNKYR
344	301	DALVLYHHINAYEEDGHVVFDIVAYRDNSLYDMFYLKKLDKDFE
366	317	:      :. .:  : ::           SPFNLFHHINTYEDNGFLIVDLCCWKGFEFVYNYFTLYLANLRENWEEVE
391		VNNKLTSIPTCKRFVVPLQYDKDAEVGSNLVKLP.TSATAVKEKDGS
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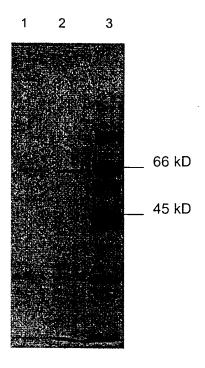


Fig. 6 shows a 10% polyacrylamide gel with E.coli expressed  $\beta,\beta$ -carotene 15,15′-monooxygenase after affinity tag purification; lane 1 and lane 2: 2 fractions from the Co²⁺-chelate column showing the main band at 60 kD; lane 3: low range molecular weight marker (Bio Rad).



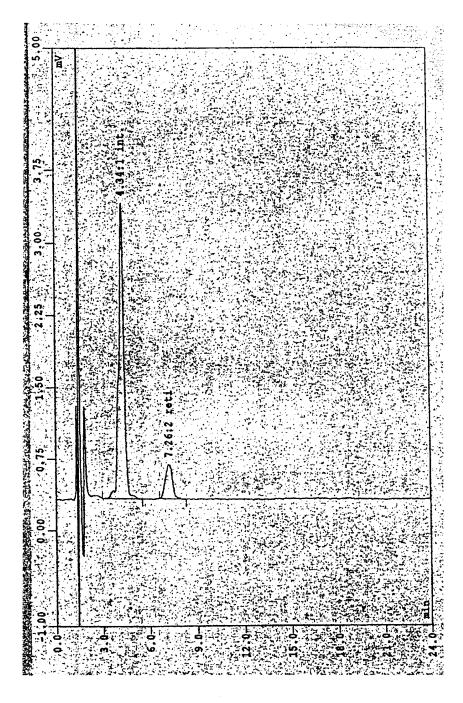


Fig. 7 shows an HPLC profile of the reaction mixture at the end of an activity assay for the  $\beta$ , $\beta$ -carotene 15,15'-monoxygenase following the procedure in example 1. The first peak in the chromatogram, represents the internal standard, while the second peak corresponds to retinal as the only product formed during the central cleavage with  $\beta$ -carotene as substrate.



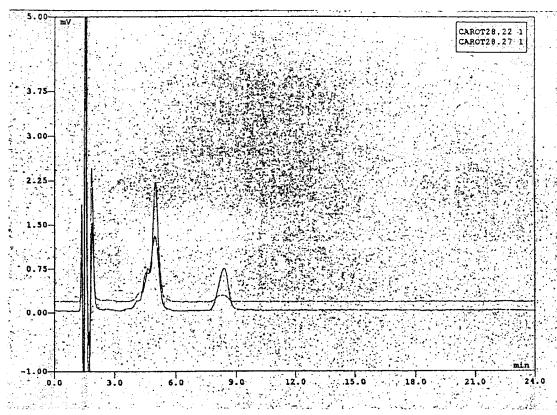


Fig. 8 confirms that the product peak in Fig. 7 is indeedretinal. A sample which was positive in the activity assay-(green (upper) chromatogram) was spiked with retinal and analysed in second HPLC run (red (lower) chromatogram). The chromatograms of the two runs were then overlayed.